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Enzymatic Deinking — A Review

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ENZYMATIC DEINKING - A REVIEW

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ABSTRACT

Waste paper recycling has increased dramatically in recent times, and will continue to do so in the foreseeable future. Ink removal constitutes one barrier to converting this raw material into quality products. Enzymatic deinking represents one approach to lowering this barrier. Results from research to date indicate that enzymes facilitate ink removal, but brightness increases vary significantly among experimental trials. Side benefits include some improvement in freeness and paper strength. The underlying mechanisms are not known, and further research is needed to clarify them as well as to demonstrate commercial utility.

KEYWORDS

Enzymes, Cellulases, Hemicellulases, Recycling, Waste Paper, Flotation, Ink.

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INTRODUCTION

Use of waste paper as raw material for papermaking has increased dramatically over the last decade. Worldwide, waste paper comprised 37 percent of the raw material supply in 1991 (1). The American Forest and Paper Association has projected that overall waste paper recovery in the USA will rise from approximately 40 percent in 1993 to 50 percent or more by 2001 (2). Profitable conversion of this relatively abundant and inexpensive raw material into quality products demands effective and efficient means for removing contaminants, with inks being one problem. Currently, 145 deinking facilities are operating, under construction, or announced for construction in the USA (3). According to Jaakko Pöyry, worldwide deinking capacity will rise to 31 million tons by 2001, with particularly large expansions for newsprint, printing, and tissue grades (4). Meeting these ambitious goals requires intensified investment in research on and development of improved deinking processes.

Some paper grades, e.g., newspapers printed with oil-based inks, can be deinked with relative ease. Even so, the best deinking processes remove only about half the ink (5). Nonimpact printed papers are more difficult to deink (6), and the quantity of such papers continues to grow as a proportion of total waste paper volume. Similarly, color printing via offset lithography is expanding in the USA at an annual rate of 25 percent (7), and the cross-linking inks used in this process are also difficult to remove. Thus, ink removal remains a major technical obstacle to greater use of recycled paper. In addition, deinking processes are substantial sources of solid and liquid waste. Disposal is a problem, and deinking plants would benefit from more effective and less polluting processes.

Enzymatic deinking may provide a means to meet these needs. Useful enzymes, e.g., cellulases, are now available in larger quantities and at lower cost than in the past (8). Enzymes have not been used for commercial deinking, but laboratory, pilot plant, and mill trials have been conducted. Results appear promising, with enzymatic treatment typically yielding residual ink areas on a par with, or better than, those produced by chemical treatment in conjunction with washing or flotation.

This paper summarizes significant findings from recent research on enzymatic deinking, documents effectiveness by enzyme and paper type, compiles information on mechanisms, and highlights research needs. Most available literature concerns cellulases and hemicellulases, and this paper therefore focuses on these enzymes. Other enzymes, e.g., lipases, are discussed only briefly.

ENZYMES AND ENZYMATIC ACTIVITIES

Deinking waste paper involves dislodging ink particles from fiber surfaces, and separating dispersed ink from fiber suspensions by washing or flotation. Enzymatic approaches involve either attacking the ink or fiber surfaces. Lipases and esterases can degrade vegetable oil-based inks. Pectinases, hemicellulases, cellulases, and lignolytic enzymes are believed to alter fiber surfaces or bonds in the vicinity of ink particles, thereby freeing ink for removal by washing or flotation. Patent claims have been filed or granted for a variety of enzymes (Table 1).

Table 1: Patents and patent applications on enzymatic deinking.

Title	Enzyme
Chemicals for Deinking (9)	N.G.
Deinking Chemicals (10)	N.G.
Enzymatic Deinking Process (11)	Alkaline Cellulase
N.G. (12)	Alkaline Cellulase
Elimination of Ink from Reclaimed Paper (13)	Alkaline Cellulase
Elimination of Ink from Reclaimed Paper (14)	Cellulase
Deinking Waste-Printed Paper Using Enzymes (15)	Cellulases
Process for Removing Printing Ink from Waste Paper (16)	Cellulase or Pectinase
Deinking of Printed Wastepaper by Cellulolytic Enzymes (17)	Cellulase or Pectinase
Biological Ink Elimination from Reclaimed Paper (18)	Cellulase and/or Pectinase
Elimination of Ink from Reclaimed Paper (19)	Esterase
Elimination of Ink from Reclaimed Paper (20)	Esterase
Process for Wastepaper Preparation with Enzymatic Printing-Ink Removal (21)	Lignolytic Enzyme (Laccase)
Deinking Waste Paper with the Incorporation of Lipase (22)	Lipase
Removal of Ink from Recycled Paper (23)	Lipase

The diversity of lignocellulolytic micro-organisms in nature serve as a rich source of enzymes for industrial applications. Such organisms, even when grown under controlled fermentation conditions, produce complex mixtures of enzymes, some of which produce unwanted side-effects. Producing specific enzymes in quantities sufficient for commercial use involves costly purification. Increased usage and advances in fermentation and purification technology are expected to lower production costs.

Alternatively, genetic engineering techniques can be used to identify the gene for a specific enzyme, and transfer it to another organism, e.g., *Escherichia coli*, that normally does not produce the enzyme. This approach was used by Paice et al. to move a β-xylosidase gene from *Bacillus subtilis* to *E. coli* (24, 25, 26). A pure xylanase was produced and used for pulp bleaching. Absence of normally associated cellulases prevented damage to the pulp. Transfer and expression of cellulase genes have also been accomplished (27), and several firms now produce individual cellulases.

Cellulases

Cellulases of fungal and bacterial origin are components of large systems or complexes that hydrolyze cellulose to water soluble sugars. Nature and action of such enzymes has been summarized by Wood and Garcia-Campayo (28), and by Eriksson (29, 30). Recent research shows that oxidative and oxidoreductive enzymes are also involved (30). Hydrolysis of crystalline cellulose thus requires at least a 3-part system, comprised of endo-1,4-β-glucanases, exo-1,4-β-glucanases, and 1,4-β-glucosidases, with each enzyme performing its particular function.

Endoglucanases hydrolyze amorphous cellulose and soluble derivatives by randomly splitting internal β-1,4-glucosidic linkages along cellulose molecules. Products include glucose, cellobiose, and other oligomers. Exoglucanases hydrolyze cellulose molecules from the nonreducing end and release glucose or cellobiose units. Glucosidases degrade cellobiose and other oligomers into glucose monomers (30, 31). Traditional distinctions among cellulases based on the aforementioned activities, though convenient, have become blurred. That is, overlapping specificities have been noted for endo- and exo-glucanases (32). In addition, synergistic interaction among them has been reported for crystalline cellulose (33), with combined activity being greater than the sum of individual activities. Synergistic effects, however, are low with amorphous and extensively hydrated cellulose, and nil with cellulose derivatives. Such observations suggest that directed mixtures of cellulases might be more effective at deinking than single enzymes or natural mixtures.

Cellulases share many features in common, including a structural and apparently bifunctional organization characterized by a central core containing a catalytic domain plus a tail having a glycosylated region (Block B) and a cellulose binding domain (Block A) (34, 35). For exoglucanases, the two domains have quite different functions, with Block B serving as a hinge linking Block A to the catalytic core. Block A functions either as an anchor or serves to relax the surface and inner structure of cellulosic fibers (36).

Separating the catalytic and binding domains produces differing effects (33). Though lacking hydrolytic activity, the binding domain adheres to fibers at surface discontinuities, penetrates these areas, and disrupts fiber structure. Further penetration exfoliates fibers, exposing cellulose chain ends and roughening surfaces. Despite low affinity for cellulose, the catalytic domain cleaves glycosidic linkages and smoothes fiber surfaces. These differing activities may generate contrasting outcomes (5), with roughening leading to improved fiber bonding and smoothing to increased freeness. Further research on enzyme structure and domains as well as differential activities is needed. Using the two domains separately or when recombined in various proportions could improve effectiveness of a variety of processes. Perhaps the reduced size and lower molecular weight of the catalytic domain will permit diffusion into and activity within fiber walls.

Hemicellulases

Naturally occurring hemicelluloses are much more variable in composition than cellulose. Degradation, partial or total, therefore requires even more complex enzyme systems. For example, complete degradation of a hemicellulose comprised largely of xylose would require a series of hydrolytic enzymes, including endo-1,4-β-xylanases, 1,4-β-xylosidases, α-glucuronidases, α-L-arabinofuran-osidases, and acetylxylanesterases. Endodxylanases are perhaps the best known of hemicellulytic enzymes, and are noted for initiating end-wise attacks on the xylan backbones of common hemicelluloses. β-xylosidases, on the other hand, convert water soluble dimers and oligomers to xylose (29). Eriksson et al. (30) provide a comprehensive review of hemicellulases.

ENZYME APPLICATIONS AND EFFECTIVENESS

Cellulases and Hemicellulases

Experience from textile manufacturing and pulp bleaching provides valuable lessons for research on enzymatic deinking. Operating environments are critical to success, and manifold variables must be optimized. These include, among others: temperature, pH, enzyme activity and dosage, reaction time, pulp consistency, and mechanical action (37, 38).

Most enzymes useful for deinking will function under present mill operating conditions (39). Effectiveness, however, is perhaps most limited by pH. Cellulases and hemicellulases vary in sensitivity to pH, with some having optimal activity in basic environments, others in neutral conditions, and still others under acidic conditions. Choosing an appropriate enzyme and maintaining proper pH during treatment determines success or failure. As an example, matching pulping environment pH to enzyme requirements yielded brightness levels above those obtained via conventional alkaline deinking (40). Deinking standard and colored newsprint with enzymes having optimal pH requirements between 4 and 5 worked well (7, 41). The initial pulp slurry had a pH of 5.5, and no adjustment was necessary. In contrast, an initial slurry of alkaline sized nonimpact printed paper had a pH of 8.5 (42) as compared to a pH of 5 to 6 optimal for enzymatic activity. Adjustment to pH 5.5 via acid addition was necessary for effective deinking. Having to adjust pH could raise operating costs and limit commercialization. However, using enzymes active in acidic environments may confer added benefits; acidic conditions lessen yellowing of derived products (43).

Proper enzyme dosage and reaction time vary with enzyme, paper, and ink type. Too much enzyme or overly long reaction times can damage fibers. Cellulases and hemicellulases, after all, evolved to degrade wood, and exploitation amounts to walking a fine line between desirable and undesirable effects. To date, enzyme dosage and treatment time has been determined via trial and error for the enzyme and environment in question. More research is needed in this critical area, and also on means for stopping reactions. Adding basic reagents to stop reactions may be counterproductive; i.e., such reagents like sodium hydroxide contribute to product yellowing.

Timing of enzyme addition is a significant concern, as past treatments seem to have been decided on largely subjective bases. Enzymes have been added before pulp disintegration (40, 43), after disintegration and during mixing (7, 41), and during pulping (42). Despite the diversity of approaches, objective comparisons cannot be made because many factors varied among experiments. A recent report (44) comparing several procedures, however, clearly demonstrated that enzyme addition during initial mixing of paper and reaction medium was most effective.

Enzymatic deinking, with or without flotation, would be especially attractive if surfactants and alkaline chemicals were not needed. Operating costs and environmental impacts would be lower. The literature shows mixed outcomes. In an early newsprint trial, acceptable results were obtained with cellulase treatment and flotation in the absence of other deinking chemicals (43). When computer printouts were used as furnish, however, the same enzyme and environment were inadequate for ink collection; foaming and collection agents were required. In another trial (39), nonimpact printed paper was deinked successfully without conventional deinking chemicals. The furnish was alkaline sized and contained calcium carbonate; adequate froth was generated during flotation. A more recent report indicates that enzymatic deinking of newspaper was best accomplished with addition of surfactants, but that other deinking chemicals were not needed (44). Enzymatic treatment produced COD loads 50 percent lower than those for conventional deinking. Most investigators, regardless of furnish or enzyme preparation, have included calcium carbonate and a surfactant as flotation aids.

Newsprint

Early enzymatic deinking research involved deinking newspaper with cellulases from *Trichoderma reesei* (43, 45, 46). Increasing pH from 4.7 to 8.0 decreased enzyme activity and reduced brightness of deinked pulp. Presoaking with enzymes before pulping appeared beneficial; a 10 min. presoak gave brighter and stronger pulp. Longer presoaking times decreased brightness, presumably due to reduced ink particle size (43, 45). The authors speculated that longer presoaking times allowed finely dispersed ink particles to readhere to fiber surfaces or to penetrate into porous parts of fibers, thereby limiting effectiveness of flotation. Soaking after pulping, but before flotation, adversely affected deinking. This result was also attributed to readherence of ink particles to fibers. This trial was among the first to demonstrate that brightness of enzymatically deinked pulp equaled that of conventionally deinked pulp.

Another trial tested effects of cellulases from *Tricoderma viride* and hemicellulases from *Aspergillus niger* (40). Brightness increased with increasing enzyme dosage, and with increasing reaction time at constant enzyme dosage. Soaking with enzymes before pulping was beneficial, but prolonged soaking reduced ink particle size, lowered flotation effectiveness, and decreased brightness. Highest brightness gains were obtained with a cellulase and hemicellulase mixture; the optimal blend gave higher brightness gains than conventional deinking.

Practicality of deinking with low pH cellulase and hemicellulase mixtures was tested with letterpress (41) and color offset printed (7) newsprint. All operations were performed at pH 5.5. Greatest brightness increases for letterpress paper were obtained with a hemicellulase preparation, primarily xylanase, but the lowest residual ink area, as measured via image analysis, was achieved with a cellulase preparation. For colored offset paper, best brightness values were obtained with a mixture of cellulases and hemicellulases. This same preparation, however, yielded the highest residual ink area, illustrating poor correlation between brightness and residual ink area. Both must be assessed to understand enzyme action and to ensure product quality. These same authors used similar enzymes to deink flexographic printed newspaper (47, 48). Enzymatic treatment and flotation removed the water-based ink with ease, resulting in brightness levels well above those obtained with conventional deinking. Inks of this type, however, are so finely divided and dispersed by conventional deinking that flotation is impaired (49). Such results suggest that enzyme treatment under acidic conditions would be best for deinking this raw material, and emphasize that deinking methods must be tailored to paper, ink, and printing type.

The phenomenon of ink particle size reduction merits further investigation. Regardless of ink type or printing process, enzymatic treatment tends to reduce ink particle size. As an example, reductions in particle size varied with pulping time in the presence of cellulases for standard newspaper, and overall reductions were greater than those noted in conventional deinking (43). Other workers (41, 50) showed reductions varying from 16 to 37 percent, depending on ink type. Credible explanations of causes have not appeared. Quantifying this effect, verifying its causes, and learning to control it remain major research issues.

Bleaching chemical requirements may be lower for enzymatic deinking. Newspaper pulps bleached after being deinked by enzymatic and conventional means had similar brightness values (43). Hydrogen peroxide had been included in the pulping as well as the bleaching step in conventional deinking, but only in the bleaching step of the enzymatic process. Enzymatically deinked pulps were thus easier to bleach and required half as much hydrogen peroxide. A similar trial with letterpress newspaper produced enzymatically deinked pulps with lower initial brightness levels than those for conventionally deinked pulps (50). Subsequent bleaching with hydrogen peroxide, however, produced similar brightness levels, with peroxide usage lowest for the enzymatic process. Brightness levels obtained from bleaching offset printed newspaper pulps after enzymatic deinking slightly exceeded those of pulps produced by conventionally deinked pulp with the same quantity of hydrogen peroxide applied during pulping (44).

Pulp Yields

Reductions in pulp yield, and accompanying release of reducing sugars, might be expected from hydrolytic activity of cellulases and hemicellulases. The dangers are fiber loss and heightened BOD in effluents. Limited information is available on yield reduction. Published data indicates that losses can be held to acceptable levels provided proper control is exercised over enzyme dosage and reaction time. As an example, reducing sugars were released during enzymatic deinking of old newspaper, but yield losses were immaterial (43). Relatively short reaction times were thought to have restricted enzyme attack to fibrils on fiber surfaces. In another trial with old newspaper, sugar release increased with enzyme dosage and reaction time (40). Yield was reduced by five percent, but freed sugars did not explain all the loss. Microfibrils freed from fibers by enzymatic activity were said to have been lost during flotation. Even so, yields from enzymatic deinking were higher than those obtained from conventional deinking.

Fiber, Pulp, and Paper Properties

Early workers were concerned that cellulases would reduce fiber lengths and adversely affect paper strength. However, Bauer-McNett classification of enzymatically deinked newspaper pulps yielded a short fiber fraction smaller than that from conventionally deinked pulps (43). Other investigators found similar trends - reduced fines content and improved drainage - when comparing pulps deinked with and without enzymes (7, 41, 47, 48, 50). In recent comparisons to conventional deinking, however, no significant effects were found on fiber length distribution, average fiber length, or mass of fiber fractions (44).

Limiting enzymatic action to removal of microfibrils is thought to remove sufficient hydrophilic material to improve drainage (51). Freeness may also be improved by enzymatic action on small colloidal particles (52, 53, 54) as well as fines (55). Low enzyme concentrations can destroy fines, but are not likely to harm intact fibers. Instead, fines and other small suspended particles, with their high surface area, are attacked preferentially. This was confirmed by silver-enhanced colloidal gold labeling and visualization via light microscopy (56). Enzyme binding may also improve freeness (55). Binding of cellulases or hemicellulases could aggregate small particles much like what occurs when polymers are used as retention aids. Hemicellulase treatment has been observed to reduce fines content without causing measurable hydrolysis.

One of the first reports on enzymatic deinking of old newspaper showed increases in tensile strength relative to conventional deinking (43). More recent trials comparing enzymatically deinked pulps from a variety of papers to those deinked in water gave similar results (7, 41, 47). A hemicellulase preparation produced the largest strength increase with the least improvement in freeness. Strength improvement was attributed to changes in hemicellulose composition and degradation of lignin-hemicellulose linkages. Lignin release following such treatment has been reported (41). On the other hand, mechanisms by which cellulase affects strength properties are far from clear. Cellulases, at high dosages, clearly reduce strength properties (51, 57, 58), and chemically pulped fibers are more severely degraded (59). Severe fiber damage can also increase fines and reduce freeness. Low enzyme dosages may affect only fiber surfaces. Partial digestion of fiber surfaces could promote fibrillation, resulting in improved bonding and stronger paper (58). Overall, the considerable strength improvements observed in deinking trials are meaningful, and seem consistent with those produced by enzymes used in secondary fiber renovation (51, 55, 60).

Nonimpact Printed Paper

Conventional chemical deinking is not an especially effective means for deinking nonimpact printed papers. Low efficiencies apparently result from strong adherence of toner ink particles to fibers. Also, ink particle sizes vary greatly, and the larger particles are difficult to separate from pulp suspensions via flotation and washing. Greater efficiencies have been projected for enzymatic deinking; e.g., a commercial cellulase preparation reduced particle sizes (43).

Investigators at the U.S.D.A. Forest Products Laboratory have compared the two methods for deinking xerographic office papers in a series of experiments (61). Treatment with a cellulase preparation gave the highest ink removal efficiency after flotation and subsequent washing (62). Combined chemical and enzyme treatments did not raise efficiency. In a later trial (42), commercial enzyme preparations with high cellulase activity or a combination of cellulases and hemicellulases also proved effective. In terms of residual ink areas, six of the seven preparations gave better results than chemical treatment. None of the enzyme preparations, however, produced higher brightness than chemical deinking, a result possibly caused by the bleaching effect of hydrogen peroxide used in the chemical process. Freeness of enzyme treated pulps was higher, but strength properties were slightly lower than those of chemically deinked pulps.

Other research indicated that treatment with a pure alkaline cellulase significantly improved brightness levels of xerographic and laser printed papers, relative to pulping in water without enzymes (39). Residual ink area was reduced by 94 percent. Enzymatic treatment changed fiber length distributions by decreasing fines content. Such results might be expected since such papers typically contain bleached softwood chemical pulp, and enzymes are more likely to affect fiber distributions of chemical pulps. Freeness was improved, and paper strength properties were either similar or slightly increased.

Effects of Additives

Direct physical contact between enzyme and substrate is prerequisite to activity (63). Paper sizing and other additives may prevent or limit contact. Implications of sizing effects, however, have been investigated only recently (64, 65, 66, 67). This first research, involving enzymatic deinking of printed cotton fabrics, suggested that sizing physically shields fibers from enzymes. Such outcomes are not surprising. Earlier work with textiles showed that starch sizing must be removed via a-amylase or other treatment before cotton fabrics can be altered by cellulase treatment (68, 69).

Paper sizings differ in mode of action, and may therefore limit contact by various means. Sizing agents may limit enzyme activity by increasing fiber hydrophobicity, physically shielding fiber surfaces from enzyme attachment, or by preventing access via covalent bonds with cellulose. Alkyl succinic anhydride, e.g., increases hydrophobicity, but also forms covalent bonds with cellulose.

Available data indicate that paper sizings reduce enzymatic deinking efficiency, and that reductions vary with sizing agent. Deinking efficiency for nonimpact printed papers (67) was lowest for papers sized with rosin/alum. Such papers had the greatest resistance to wetting and the highest fiber hydrophobicity. Papers sized with alkyl succinic anhydride were less resistant to wetting, but were almost as difficult to deink. Such findings affirm the need for additional research, but future investigations must not be limited to sizing effects. That is, numerous additives are used in papermaking and effects may dramatically vary. Coatings, dyes, metals, and other additives may denature or inhibit enzymes.

Other Approaches to Enzymatic Deinking

Another enzymatic approach to deinking involves attacking ink. Much research has been done on this front, and several patents claim that alkaline lipases facilitate removal of oil-based inks (Table 1). Published data (70) indicates that an alkaline lipase efficiently removed offset printing ink. The effect was attributed to enzymatic hydrolysis of drying oil or thermosetting resin in the ink. Lipases should be effective with inks carried in natural vegetable oils, and the approach merits additional research, especially if the trend toward greater utilization of such inks accelerates.

The high proportion of lignin-rich mechanical pulps in newsprint suggests that enzymes catalyzing removal of surface lignin may hold promise for deinking. This approach has been evaluated using an intact organism (*Phanerochaete chrysosporium*) (71) and with lignin degrading enzymes (72, 73). Lignin release was observed in all cases. Ink removal by a laccase preparation proved comparable to that of conventional chemical deinking (72, 73). Pulps deinked with laccase showed high brightness, and were easier to bleach. Further research on such enzymes, used alone, or in combination with cellulases and/or hemicellulases, could lead to an improved understanding of the interactions between fiber surfaces and ink, if not improved deinking effectiveness.

Recent literature contains a report of a novel deinking process coupling separation technology with cellulase treatment (74, 75). Ink particles dislodged from newsprint, presumably by cellulase activity, readhered to smaller fibers originally present or created by enzymatic action. The smaller fibers and adhered ink, were then separated from longer, deinked fibers. The latter were considered useable without further treatment. Since ink adhered to the shorter fibers, conventional washing or flotation would be unnecessary, and ink would not be released into effluents. Cause of the strong association between ink and short fibers was not identified. That such separation may be practical, however, has been shown (76, 77).

POSSIBLE MECHANISMS

A variety of mechanisms have been proposed for removal of ink by enzymes. The role of lipases and esterases, of course, is understood; such enzymes degrade carrier oils and disperse pigments. The situation is not nearly so clear for hydrolytic enzymes. Most authors have advanced at least general explanations, but few have supplied definitive supporting data. Thus, the nature of the underlying causes remains uncertain. That mechanisms have not been definitively elucidated should not be surprising. Research on this promising technology has been underway for a relatively short time, and discerning mechanisms is more difficult than demonstrating effectiveness. Also, the diversity of paper grades and printing processes make it unlikely that one or only a few mechanisms are involved. A short summary of the most frequently advanced explanations follows:

1) Deinking may be caused not by enzymes but by additives used to enhance enzyme production and stabilization. Residual ink areas obtained with heat-deactivated enzymes, however, did not exceed those observed after pulping in water (42). Also, ink removal varied inversely with enzyme inhibition (64, 65, 66).

2) Catalytic hydrolysis may not be essential; enzymes can remove ink under nonoptimal conditions. Mere cellulase binding may disrupt fiber surfaces in a manner and to an extent sufficient to release ink during pulping (75).

3) Enzymes partially hydrolyze and depolymerize cellulose molecules at fiber surfaces, thereby weakening bonds between fibers and freeing them from one another. Ink particles simply are dislodged as fibers separate during pulping (43).

4) Enzymatic treatment weakens bonds, perhaps by increasing fibrillation or removing surface layers of individual fibers (16). The suggestion that enzymatic activity could be sufficient to remove whole surface layers at the low dosages and short reaction times commonly employed, however, is questionable.

5) Hemicellulases facilitate deinking by breaking lignin-carbohydrate complexes and releasing lignin from fiber surfaces (40). Ink particles are dispersed with the lignin. Cellulase and hemicellulase treatment facilitated ink removal from newsprint, and was accompanied by release of lignin (41, 47).

6) Cellulases peel fibrils from fiber surfaces thereby freeing ink particles for dispersal in suspension (16). This peeling mechanism (78, 79) has also been implicated in pulp freeness increases after enzymatic treatment of secondary fiber (51, 55). Enzyme dosages and reaction times, however, seem too low to cause measurable cellulose degradation (42).

7) Mechanical action is critical and prerequisite to enzymatic activity (64, 65, 66). Experiments involving mechanical and enzymatic treatment of printed cotton and rayon fabric showed that deinking efficiency increased linearly with applied friction. Results agreed with those from combined enzymatic treatment and stone washing of textiles. Mechanical action was said to distort cellulose chains at or near fiber surfaces, thereby increasing vulnerability to enzymatic attack. Assuming that fiber-fiber friction increases with pulp consistency, such an explanation seems consistent with earlier findings that enzymatic deinking is more effective at medium consistency as opposed to low consistency (42). Other research, however, disputes the importance of mechanical action (44). Applying greater shear forces via pulping at higher consistencies or for extended times did not improve brightness. Also, high shear forces caused by fiber-fiber friction can denature enzymes (80).

8) Enzymatic effects may be indirect, i.e., they remove microfibrils and fines, thereby improving freeness and facilitating washing or flotation (42). Fines content, however, is not always reduced during enzymatic deinking (44).

9) Enzymatic treatment of nonimpact printed paper removed fibrous material from ink particles, increasing particle hydrophobicity and facilitating separation during flotation (42). This promising hypothesis should be tested with a wide array of enzymes, paper grades, and inks.

CONCLUSIONS

Results from enzymatic deinking trials generally appear promising, but further research is needed to determine if their use offers significant cost advantages over conventional methods. Such research will not advance rapidly, however, unless substantial effort is also committed to clarifying underlying mechanisms.

Enzymatic treatment with subsequent flotation and/or washing typically results in residual ink areas significantly lower than those produced by chemical treatment in conjunction with washing or flotation. Results in terms of brightness have been mixed, with enzymatic treatment often giving brightness values less than or only on a par with conventional deinking. This differential outcome has been observed repeatedly, and may result from the tendency of enzymes to reduce ink particles to much smaller sizes than other methods. This effect, an advantage in deinking nonimpact printed papers, could present a serious obstacle to the commercial deinking of other papers. Reduced particle sizes can limit flotation efficiency. Should particles be fine enough to diffuse into fibers, even washing efficiency could be reduced. Until this phenomenon is understood, enzymatic deinking will have limited commercial potential.

Continued increases in use of nonimpact printed papers, coupled with the effectiveness of enzymes in deinking this type of paper, may mean that first commercial usage of enzymatic deinking will be with this type of paper. Changes in printing processes will undoubtedly affect deinking technologies in the future. Enzymes that attack inks rather than fibers could, therefore, become more useful.

A variety of enzymes and mixtures are now available. As a result, deinking should be possible in whatever pH environment is encountered. Best results to date typically have been obtained with mixtures containing a cellulase or cellulases and small amounts of hemicellulase. Eventual availability of individual cellulases and/or hemicellulases will permit tailoring treatment to paper grade and printing process, while maintaining balance between effects on ink removal and fiber properties. Intensified research on enzyme structure and function will hasten developments in this area.

The various additives used in papermaking may limit effectiveness of enzymatic deinking. Sizing agents can physically prevent access to cellulose. Little is known about the effects of additives, and the potential for reduced effectiveness makes this a prime area for research.

While the picture is not clear, evidence is accumulating that enzymes used in deinking also produce positive changes in pulp and paper properties, e.g., improved freeness and increased tensile strength. This can be expected to yield important cost savings and quality improvements. Prospective users of hydrolytic enzymes will remain skeptical, however, until enzyme-fiber interactions have been clarified. One prominent concern is the differential effect of enzymatic treatment on chemical, mechanical, and secondary fibers. Opportunities to apply findings from deinking research to improving secondary fiber properties and the reverse should not be overlooked.

Enzymatic deinking may yield a number of side benefits. The feasibility of deinking in acidic environments has been confirmed. Applied commercially, this should reduce overall chemical requirements and minimize yellowing of reclaimed papers after alkaline deinking. Reduced chemical usage means lower waste treatment costs and less impact on the environment. Lower bleaching costs can also be anticipated; enzymatically deinked pulps have proven to be easier to bleach and require less bleaching chemicals than pulps deinked by conventional means.

Few cost comparisons between enzymatic and conventional deinking have been published. In trials using typical commercial cellulases to deink nonimpact printed papers, enzyme costs ranged from $0.40 to 2.40/1000 kg paper (42, 62). Some added costs, though not documented, were incurred to adjust pH levels. Other process steps were considered to cost the same as those in the conventional procedure. Chemical costs for the conventional process approached $20/ 1000 kg; costs of the two methods were therefore considered similar. Such analyses were based on small scale laboratory tests; larger scale trials are needed for thorough evaluation.

Future economic analyses should also consider other potential benefits associated with enzyme usage, e.g., lower energy consumption, reduced chemical needs for ink removal and flotation, and lower bleaching requirements. Improved drainage and faster machine speeds resulting from freeness increases may also yield significant cost savings. Similarly, enzymatic deinking is likely to produce simpler and fewer waste disposal problems. Some capital savings could also accrue as a result of more efficient dispersion and flotation. These benefits will be important only if the deinking efficiencies described in the literature are realized.

Finally, enzyme costs can be expected to fall in the future as demand rises. Genetic engineering can be expected to simplify production and lessen purification costs. The future may also see development of biomimetic catalysts. These compounds are simpler and have lower molecular weights than enzymes, but maintain function and specificity. Such synthetic catalysts would be more stable in commercial deinking environments, thereby permitting wider application.

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